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Claims

1. A method for determination of amounts or relative proportions of more than one individual polynucleotide sequence or subgroups thereof in a polynucleotide mixture using a quantitative affinity aided solution hybridization in combination with size- or mass-based fractionation for obtaining resolution, **characterized** in that the method comprises the consecutive steps of:
 - (a) providing, one or more organized pools with a preset optional number of soluble polynucleotide probes, each probe being complementary to an individual target ribopolynucleotides in the sample, being present in a molar excess as compared to the analyte polynucleotide sequences, and having approximately the same number of hybridizing nucleotides, which probes are made distinguishable by providing said polynucleotide probes with one or more resolution enabling tags, which change the size or mass and thereby provide the polynucleotide probes with different mobilities in fractionation, separation or recording systems without disturbing the hybridization or capturing reaction;
 - (b) providing the analyte polynucleotide sequences isolated from a sample comprising a mixture of target ribopolynucleotide sequences with at least one affinity tag; and thereafter
 - (c) performing steps (i) and (ii) simultaneously, or sequentially; in the order (i) and (ii), wherein the steps (i) and (ii) comprises
 - (i) allowing a hybridization reaction to take place between the molar excess of soluble polynucleotide probes from step (a) and the analyte ribopolynucleotide sequences from step (b) leading to a quantitative formation of soluble hybrids;
 - (ii) recovering the hybrids, which have been quantitatively formed in step (i) by capturing said hybrids quantitatively on a separation aiding tool provided with the affinity pair of the affinity tag of the analyte polynucleotides;
 - (d) quantitatively releasing the polynucleotide probes in an unmodified form from the hybrids captured to separation aiding tool;
 - (e) separating and recording the amount or relative proportions of distinguishable probes, the amount of which corresponds to the amount of complementary target ribopolynucleotide sequences in the mixture of analyte ribopolynucleotides in the sample.

2. The method according to claim 1, **characterized** in that for the determination of dynamic variations in the amounts or relative proportions of polynucleotide transcripts or their subgroups in an individual organism, the soluble polynucleotide probes are designed from species or group-specific ribopolynucleotide sequences hybridizing with selected more or less conserved or hypervariable regions from intragenomic sequences specific for subgroups, species, subspecies of transcripts expressed in the organism.
3. The method according to claim 2, **characterized** in that the analyte polynucleotide sequences isolated from the sample comprising the mixed target population is messenger RNA (mRNA).
4. The method according to claim 1, **characterized** in that for the determination of dynamic variations in the amounts or relative proportions of polynucleotide sequences representing individual organisms or subpopulations thereof in a target population, the soluble deoxyribopolynucleotide probes are designed from species or group-specific ribopolynucleotide sequences hybridizing with a selected more or less conserved or hypervariable region from intragenomic sequences specific for and/or representing different phylogenetic levels allowing the identification of subgroups, species, subspecies within the mixed target population.
5. The method according to claim 4, **characterized** in that the ribopolynucleotide analytes isolated from the sample comprising the mixed target population comprise ribosomal RNA.
6. The method according to claim 1, **characterized** in that the resolution enabling tag may simultaneously act as a tracer, affinity and/or primer tag.
7. The method according to claim 1, **characterized** in that the resolution enabling tag is separatable in a sieving medium.
8. The method according to claim 7, **characterized** in that the resolution enabling tag which additionally may act as an affinity and/or primer tag is an oligonucleotide residue.
9. The method according to claim 7, **characterized** in that the resolution enabling tag which additionally may act as an affinity tag and/or a tracer tag is an amino acid or a peptide.

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10. The method according to claim 7, **characterized** in that the resolution enabling tag which additionally may act as a tracer tag is selected from a group consisting of labels recordable by fluorescence, luminescence, infrared absorption, electromagnetic properties, radioactivity and enzymatic activity.
11. The method according to claim 1, **characterized** in that the preset optional number of soluble polynucleotide probes in the pool is more than one preferably more than five, most preferably more than ten.
12. The method according to claim 1, **characterized** in that the amount of the individual, quantitatively captured and released polynucleotide probes is recorded with a fully or partly automated recording system, which is selected based on the applied resolution enabling tags.
13. The method according to claim 12, **characterized** in that the recording system is selected based on resolution enabling tags and comprises mass spectrometry, electrophoretic or chromatographic techniques.
14. The method according to any of claims 1-13, **characterized** in that the amount of the quantitatively recovered primer tagged probes are released and subsequently amplified and optionally tracer tagged before, during or after the PCR-reaction and thereafter recorded with a recording system selected based on the resolution enabling tags.
15. The method according to claim 14, **characterized** in that the primer is a universal primer.
16. The method according to claim 1, **characterized** in that the polynucleotide probes are stable DNA fragments, synthetic or recombinant polynucleotide sequences or modified polynucleotide sequences.
17. The method according to claim 1, **characterized** in that a comparative, quantitative assessment of variations in the amounts of individual polynucleotide sequences or organisms and subgroups thereof in a population or mixture of polynucleotide sequences by providing a set of multiple test kits, at least one test kit for each sample to be compared,

wherein each of said test kits are provided with one or more identical organized pools with a preset optional number of soluble polynucleotide probes, each probe being DNA complementary to an individual target ribopolynucleotide sequence in the sample, being present in a molar excess as compared to the target polynucleotides in the samples, and having an indistinguishable number of hybridizing nucleotides, which probes are made distinguishable by providing each polynucleotide probe with one or more resolution enabling tags, which change the size or mass and thereby provide the polynucleotide probes with different mobilities in the fractionation, separation or recording systems without disturbing the hybridization or capturing reaction, each pool of polynucleotides probes being placed in an organized manner in their own vessels, which are separate or joined together.

18. The method according to claim 17, **characterized** in that the individual test kits, wherein the resolution enabling tag is not a tracer tag, a set of multiple test kits is provided with tracer tags each being distinguishable from the other by the emitted signal.
19. The use in any method according to claims 1-18 of a test kit, **characterized** in that the test kit comprises one or more organized pools, with a preset optional number of soluble polynucleotide probes, each probe being complementary to an individual target ribopolynucleotide sequence in the sample, being present in a molar excess as compared to the analyte ribopolynucleotides in the samples, and having approximately the same number of hybridizing nucleotides, which probes are made distinguishable by providing each polynucleotide probes with one or more resolution enabling tags, which change the size or mass and thereby provide the polynucleotide probes with different mobilities in the fractionation, separation or recording systems without disturbing the hybridization or capturing reaction, each pool of polynucleotides probes being placed in an organized manner in their own vessels, which are separate or joined together.
20. The use according to claim 19, **characterized** in that for the determination of dynamic variations in the amounts or relative proportions of polynucleotide transcripts or their subgroups in an individual organism, the soluble polynucleotide probes are designed from species or group-specific polynucleotide sequences hybridizing with selected more or less conserved or hypervariable region from intragenomic sequences specific for subgroups, species, subspecies of transcripts expressed in the organism.

21. The use according to claim 19, **characterized** in that the polynucleotide analytes isolated from the sample comprising the mixed target population comprise messenger RNA (mRNA).
22. The use according to claim 19, **characterized** in that for the determination of dynamic variations in the amounts or relative proportions of polynucleotide sequences representing individual organisms or subpopulations thereof in a target population, the soluble polynucleotide probes are designed from species or group-specific polynucleotide sequences hybridizing with selected more or less conserved or hypervariable region from intragenomic sequences specific for and/or representing different phylogenetic levels allowing the identification of subgroups, species, subspecies within the mixed target population.
23. The use according to claim 22, **characterized** in that the polynucleotide analytes isolated from the sample comprising the mixed target population comprise ribosomal RNA.
24. The use according to claim 19, **characterized** in that the resolution enabling tag may simultaneously act as a tracer, affinity or primer tag.
25. The use according to claim 24, **characterized** in that the resolution enabling tag is separable in a sieving medium.
26. The use according to claim 25, **characterized** in that the resolution enabling tag which additionally may act as an affinity tag and/or primer tag is an oligonucleotide residue.
27. The use according to claim 25, **characterized** in that the resolution enabling tag which additionally may act as an affinity tag and/or a tracer tag is an amino acid or a peptide.
28. The use according to claim 25, **characterized** in that the resolution enabling tag which additionally may act as a tracer tag is selected from a group consisting of labels recordable by fluorescence, luminescence, infrared absorption, electromagnetic properties, radioactivity and enzymatic activity.

29. The use according to claim 19, **characterized** in that the preset optional number of soluble polynucleotide probes in the pool is more than one preferably more than five, most preferably more than ten.
30. The use according to claim 19, **characterized** in that the soluble pools of polynucleotide probes are placed in wells on a microtiter plate.
31. The use according to claim 19, **characterized** in that the polynucleotide probes are stable DNA fragments, synthetic, recombinant or modified polynucleotide sequences.
32. The use according to claim 19, **characterized** in that for a comparative, quantitative assessment of variations in the amounts of individual polynucleotide sequences or organisms and subgroups thereof in a population or mixture of polynucleotide sequences comprises a set of test kits, wherein at least one identical test kit having identical pools of polynucleotide probes for each sample to be compared.
33. The use according to claim 32, **characterized** in that each individual test kits in the set of multiple test kits is provided with tracer tags, which are distinguishable from each other by the emitted signal.
34. Use according to claim 32 for assessing hygienic conditions and epidemiologic situations, effects of external stimuli or treatment modalities on a microbial population.
35. The use according to claim 34, wherein the external stimulus or treatment is selected from a group consisting of treatment with antibiotics or hygienic measures.